A Stochastic Model of Pulsatile Blood Testosterone Levels

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It has been observed that blood testosterone levels in men oscillate with a period of 2 to 3 hours. One simple, deterministic model was proposed to describe this phenomenon, but it was recently proved that the model has a globally stable fixed point. Therefore, the deterministic model cannot observe oscillations. We will take a closer look at this model from a different physical basis in which intrinsic fluctuations are considered. It turns out that sustained oscillations do arise in the continuous-time, discrete-state stochastic model.

Outline

- Overview of the hypothalmus-pituitary-testicular axis.
- Introduce the deterministic model.
- Reconsider the model as a stochastic model.
  - Incorporate intrinsic fluctuations.
  - The Gillespie Algorithm.
- Analysis and discussion.
- Conclusions and future work.

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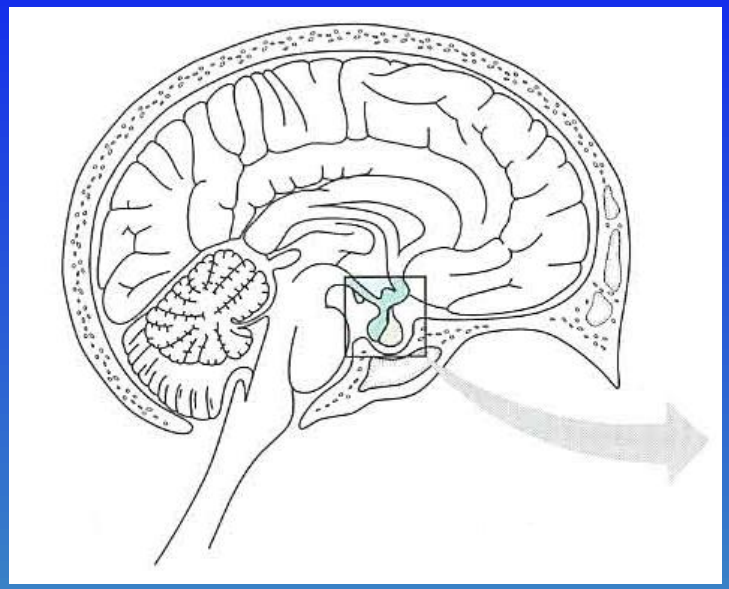
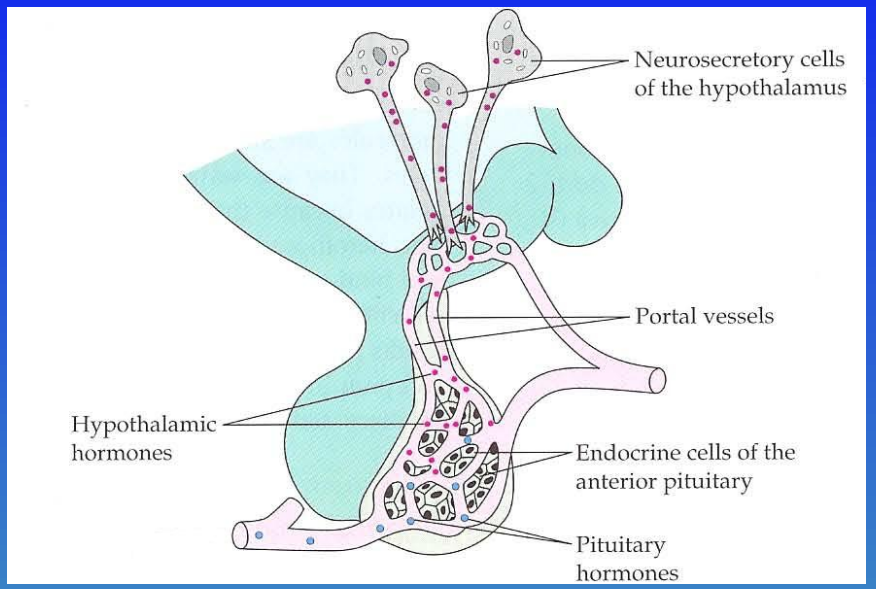
- An imbalance can cause dramatic changes (mood, acne, and weight).
- Pathway is associated with many other important processes in the body.
- Pharmaceutical interests in chemical castration (Goserelin, Lupron, and Depo-provera) and the creation of a male *pill*.

The Hypothalmus-Pituitary-Testicular Axis





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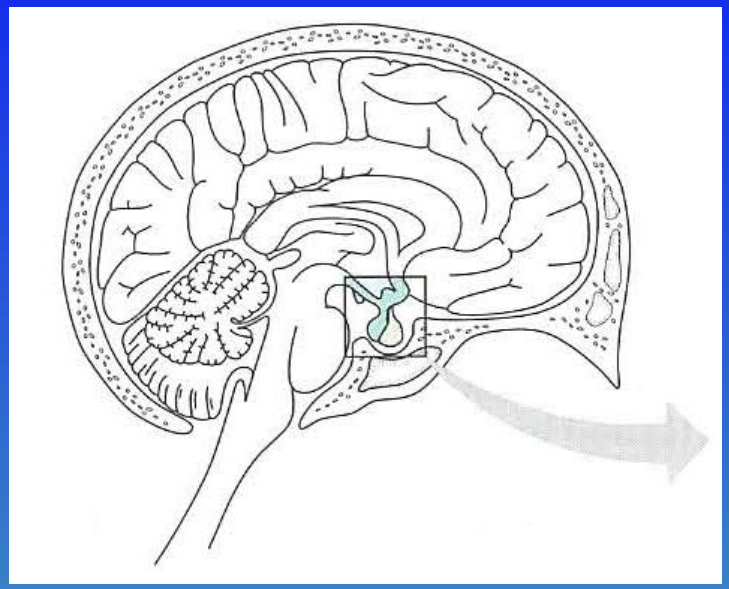
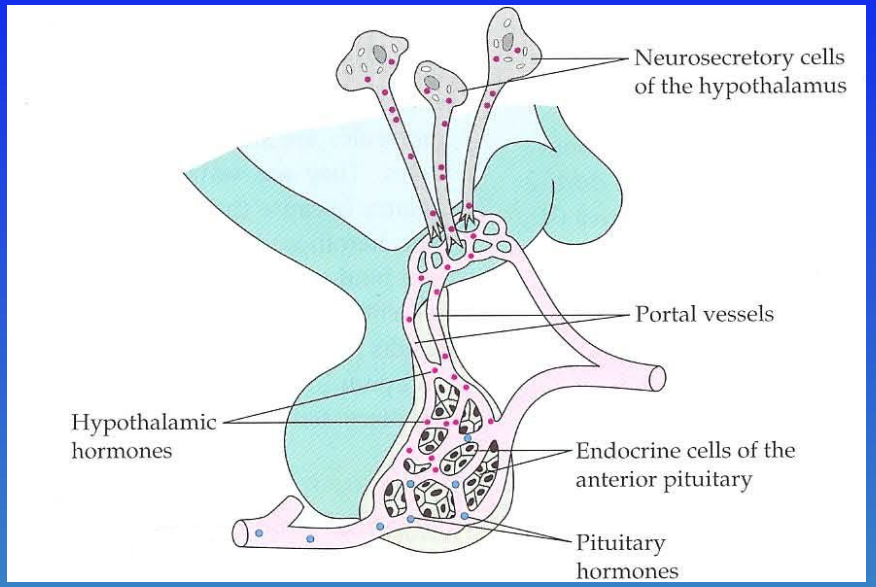
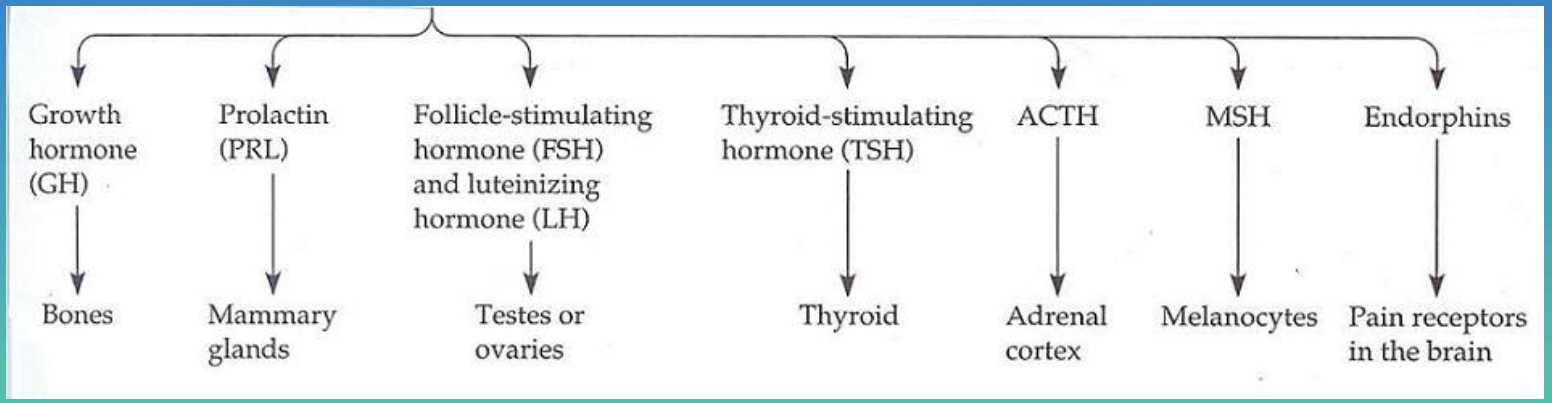
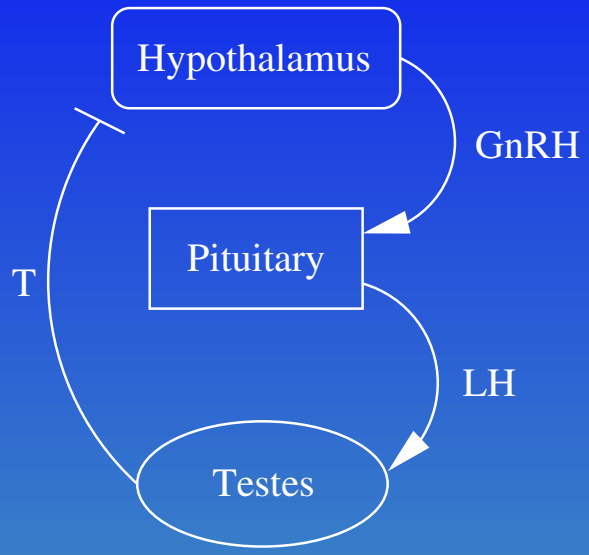






Modified from Campbell (1996).

The Hormone Secretion Signaling System

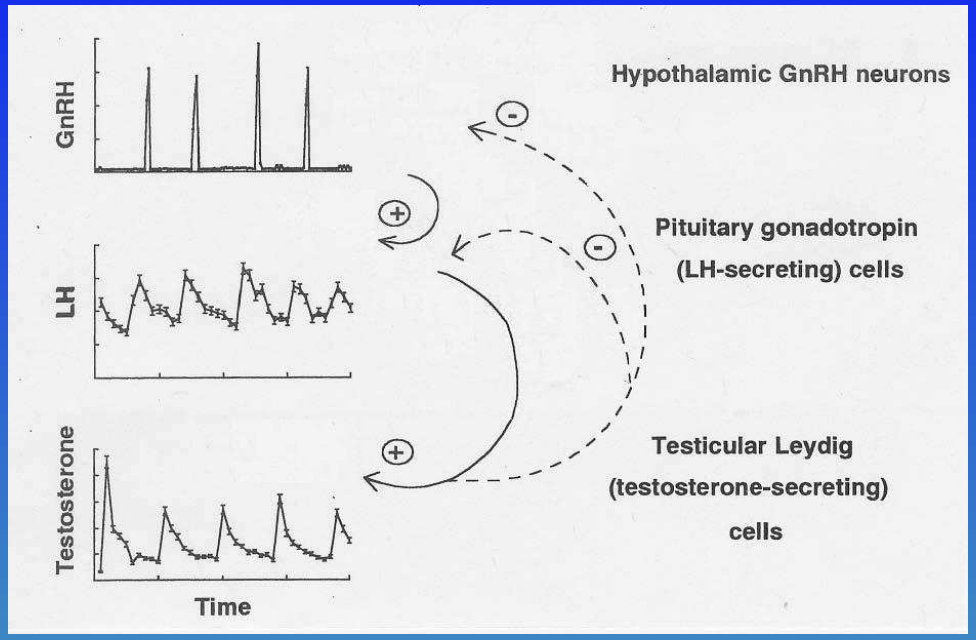


$GnRH =$ Gonadotropin Releasing Hormone

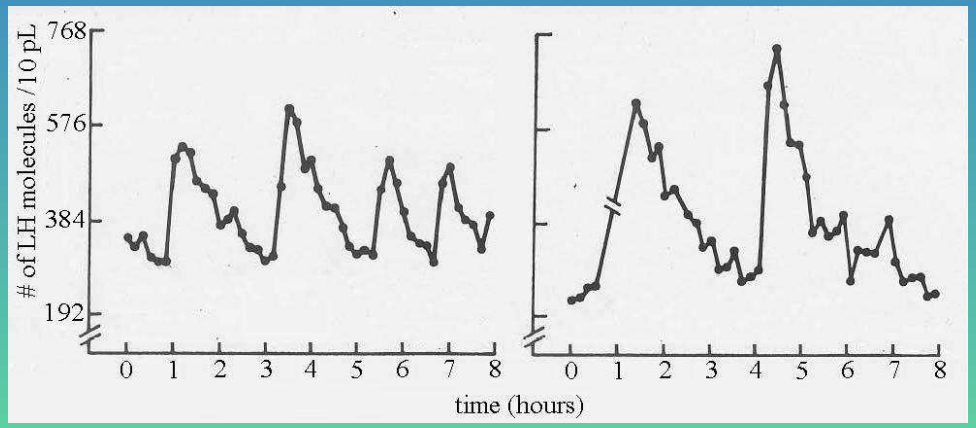
$LH =$ Luteinizing Hormone

$T =$ Testosterone

Experimental Observations



Modified from Yen *et al.* (1999).



Modified from Naftolin *et al.* (1973).

The Deterministic Model

If we represent the concentrations of *GnRH, LH*, and *T* by *R(t), L(t)*, and *T(t)*, respectively, then a proposed deterministic model of this system is

$$\frac{dR}{dt} = f(T) - b_1 R$$
$$\frac{dL}{dt} = g_1 R - b_2 L$$
$$\frac{dT}{dt} = g_2 L - b_3 T$$

where

$$f(T) = \frac{A}{K + T}$$

and A, K, b_1, b_2, b_3, g_1, and g_2 are all positive constants.

History of the Model

- Goodwin (1964) first proposed the model to demonstrate oscillatory behavior in enzymatic control processes.
- Smith (1980) studied slight variation involving a Hill coefficient in $f(T)$.
- Murray (1989) suggested using a time-delay in the production rate of T.
- Enciso and Sontag (2004) proved that the system has a globally stable fixed point (regardless of the length of the time-delay) and therefore does not have a limit cycle or sustained oscillations.
- More detailed (and more complicated) models include those by Cartwright and Husain (1986) and Keenan *et al.* (1998 and 2000).

The Fixed Point

The system of differential equations has a fixed point wherever

$$R^* = \frac{1}{b_1} f(T^*)$$

$$L^* = \frac{g_1}{b_2} R^*$$

$$T^* = \frac{g_2}{b_3} L^*$$

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or, plugging in the assumed form of $f(T)$ and solving, we find a positive fixed point at

$$R^* = \frac{-Kb_1b_2b_3 + \sqrt{(Kb_1b_2b_3)^2 + 4b_1b_2b_3g_1g_2A}}{2b_1g_1g_2}$$
$$L^* = \frac{-Kb_1b_2b_3 + \sqrt{(Kb_1b_2b_3)^2 + 4b_1b_2b_3g_1g_2A}}{2b_1b_2g_2}$$
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Stability of the Fixed Point

LOCALLY:

The characteristic equation of the linearized system near the fixed point is

$$(\lambda + b_1)(\lambda + b_2)(\lambda + b_3) - f'(T^*)g_1g_2 = 0$$

which only has solutions with negative real parts, i.e. $Re(\lambda) < 0$.

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SO:

The deterministic model does not have limit cycles and cannot have sustained oscillations, which was the purpose of the model! So what do we do next?

Reconsider the Physical Basis of the Problem

Take seriously the fact that events, such as the production or degradation of hormone molecules, occur in an essentially random manner. Intrinsic fluctuations play an important role when there are low numbers of molecules present.

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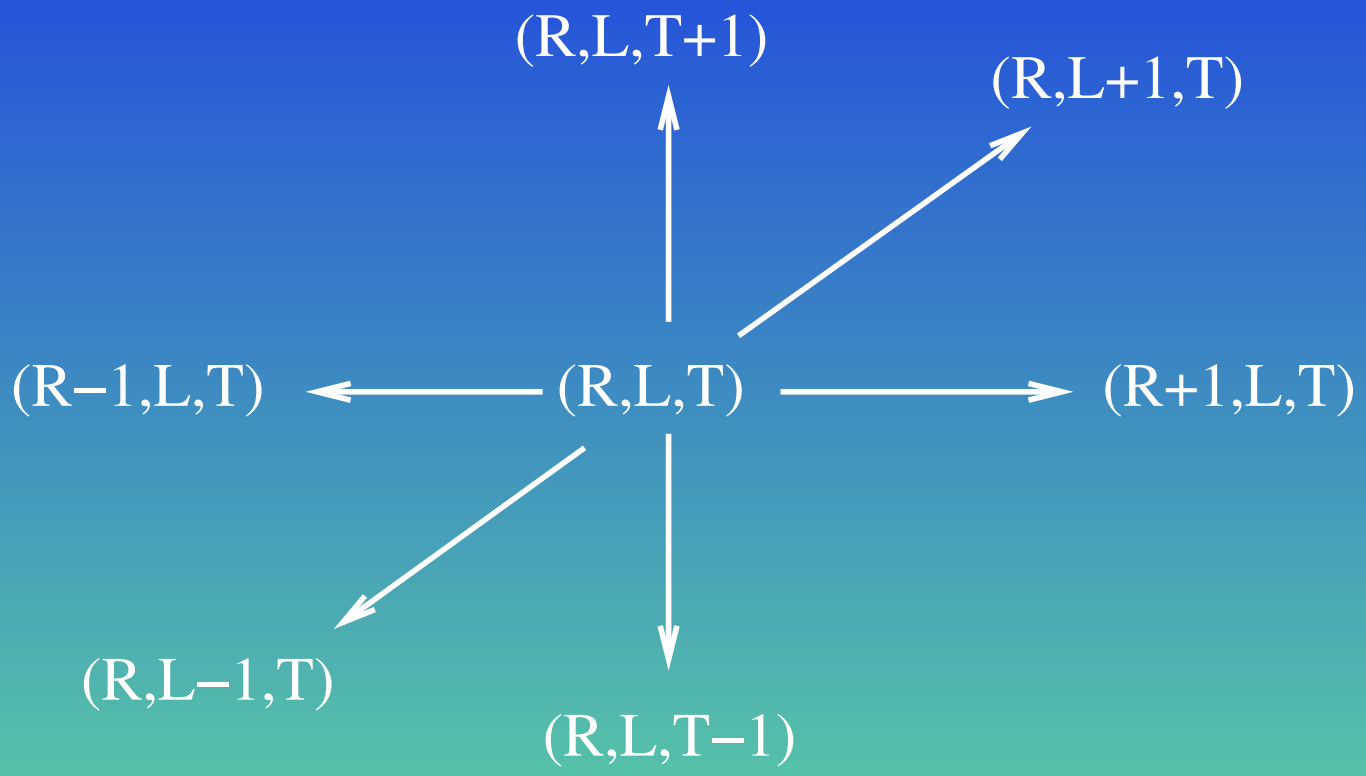

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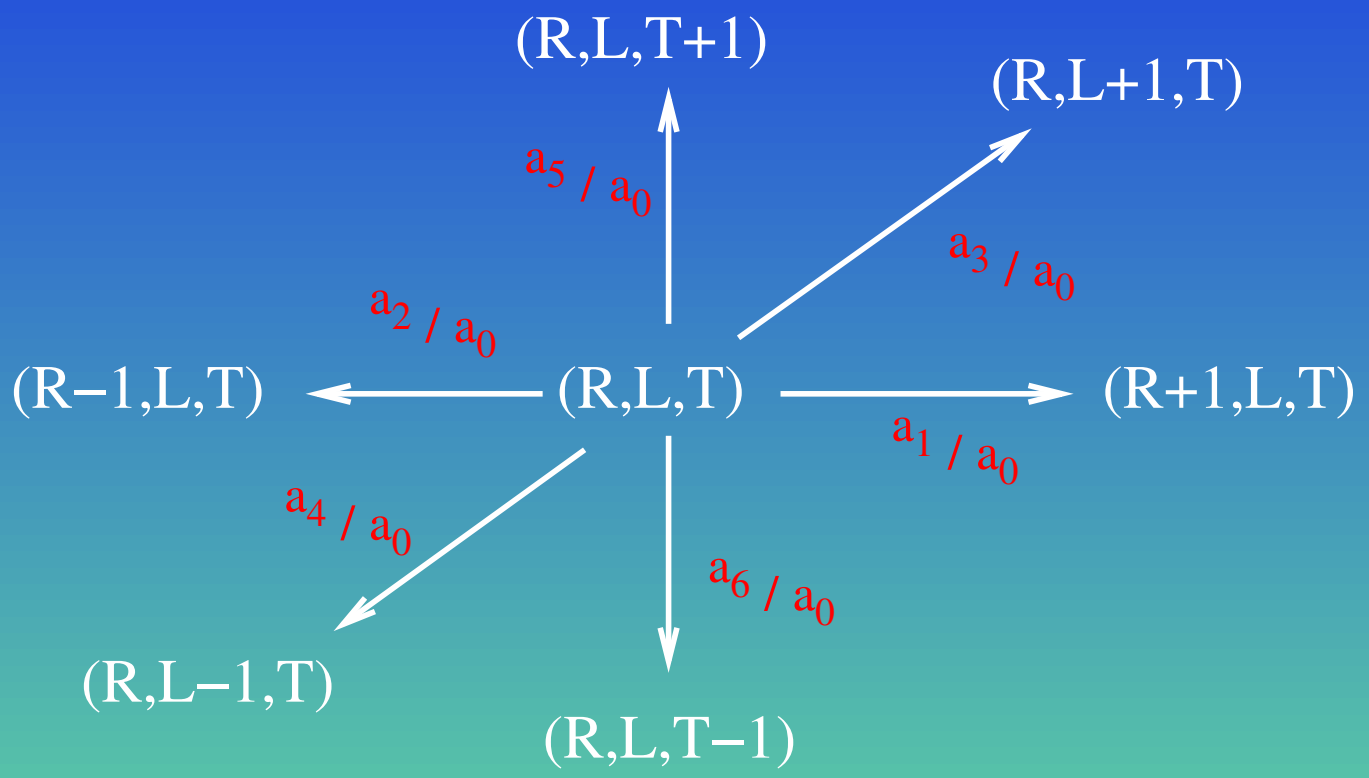

Modeling Approaches

Three types of modeling regimes: discrete and stochastic, continuous and stochastic, and continuous and deterministic regimes.

- Deterministic: the law of mass action.
- Stochastic: the chemical master equation.

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Deterministic models are the infinite volume limit of the Markov chain models.

- Kurtz, *J. Chem. Phys.* (1972).

Stochastic Simulations

- Most biochemical networks are very complex and it is not possible to obtain analytic solutions when modeling them. For this reason, we turn to stochastic simulation algorithms.

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- Gillespie (1976) developed an influential, exact method for simulating these networks.
- Other approximation methods have been developed to streamline the simulations and reduce computational overhead.

The Chemical Master Equation

The master equation is the time-evolution equation for the function $P(\mathbf{n}, t)$, where n_i is the number of molecules of species X_i in a well-mixed volume.

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$$P(\mathbf{n}, t+dt) = P(\mathbf{n}, t)P(\text{there is no change within } dt)$$
$$+ \sum_{\mu=1}^{M} P(\mathbf{n}-\mathbf{s}_\mu, t)P(\text{reaction } \mu \text{ occurs within } dt),$$
$$= P(\mathbf{n}, t)\left(1 - \sum_{\mu=1}^{M} a_\mu(\mathbf{n})dt\right) + \sum_{\mu=1}^{M} P(\mathbf{n}-\mathbf{s}_\mu, t)a_\mu(\mathbf{n}-\mathbf{s}_\mu)dt,$$

where $a_\mu(\mathbf{n})dt$ is the probability that reaction μ will occur in $(t, t+dt)$ given that the system is in state $\mathbf{n}$ at time t and $\mathbf{s}_\mu$ is a stoichiometric vector defining the result of reaction μ.

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where $a_\mu(\mathbf{n})dt$ is the probability that reaction μ will occur in $(t, t+dt)$ given that the system is in state $\mathbf{n}$ at time t and $\mathbf{s}_\mu$ is a stoichiometric vector defining the result of reaction μ. From this we get

$$\frac{dP(\mathbf{n}, t)}{dt} = \sum_{\mu=1}^{M} a_\mu(\mathbf{n}-\mathbf{s}_\mu)P(\mathbf{n}-\mathbf{s}_\mu, t) - a_\mu(\mathbf{n})P(\mathbf{n}, t).$$

The Reaction Probability Density Function

So we ask: “When will the next event occur and what type of event will it be?”

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Define

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This joint probability density function can be written as

$$P(\tau,\mu)d\tau = P_0(\tau)a_\mu d\tau = a_0 e^{-a_0\tau}\left(\frac{a_\mu}{a_0}\right)d\tau = P(\tau)P(\mu)d\tau$$

where $P_0(\tau)$ is the probability that no event occurs in the time interval $(t, t+\tau)$.

Density Function Cont'd...

Let

$$a_0 = \sum_{i=1}^{6} a_i$$

then we have

$$P_0(t + dt) = P_0(t)(1 - a_0 dt)$$

or, rearranging a little, we have

$$\frac{P_0(t + dt) - P_0(t)}{dt} = -a_0 P_0(t)$$

from which it is easily deduced that

$$P_0(t) = e^{-a_0 t}.$$

Density Function Cont'd...

So we have

$$\begin{aligned} P(\tau,\mu)d\tau &= P_0(\tau)a_\mu d\tau \\ &= \left(\frac{a_\mu}{a_0}\right) a_0 e^{-a_0\tau} d\tau \\ &= P(\mu)P(\tau)d\tau \end{aligned}$$

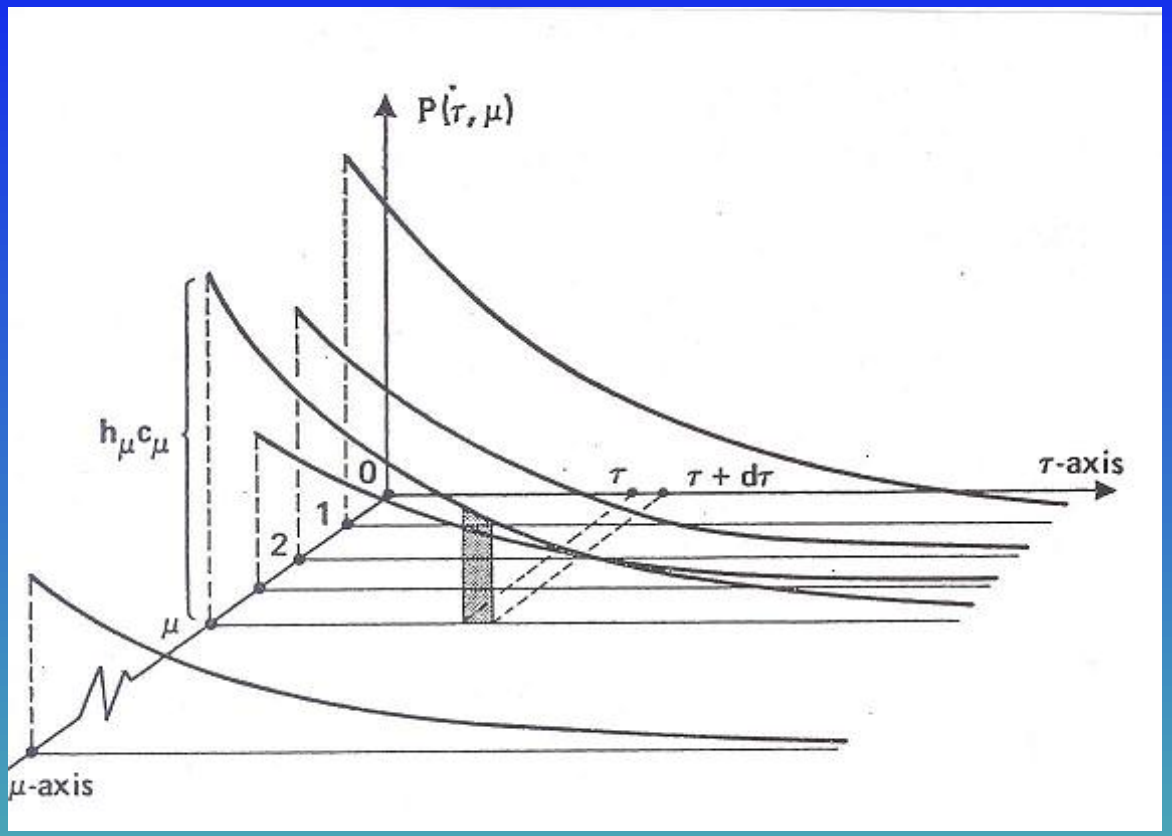
where

$$P(\mu) = \frac{a_\mu}{a_0}$$

and

$$P(\tau) = a_0 e^{-a_0\tau}.$$

Schematic of the Density Function



(Gillespie, 1976)

The Probability Distribution Function

$$F(x) \equiv \int_{-\infty}^{x} P(x')dx'$$

To generate a random value x according to a given density function $P(x)$ we need to use the inversion method, by which we simply draw a random number r from the uniform distribution in the unit interval and take x such that

$$F(x) = r \quad \text{or} \quad x = F^{-1}(r)$$

since

$$F(x' + dx') - F(x') = F'(x')dx' = P(x')dx'.$$

The Gillespie Algorithm

$$P(\tau) = a_0 e^{-a_0 \tau} \quad \longrightarrow \quad F(\tau) = 1 - e^{-a_0 \tau}$$

$$P(\mu) = \frac{a_\mu}{a_0} \quad \longrightarrow \quad F(\mu) = \sum_{k=1}^{\mu} P(k)$$

So choose r_1 and r_2 from uniform distribution in the unit interval and

$$\tau = \frac{1}{a_0} \ln\left(\frac{1}{r_1}\right)$$

$$\sum_{k=1}^{\mu-1} \frac{a_k}{a_0} < r_2 \leq \sum_{k=1}^{\mu} \frac{a_k}{a_0}.$$

The Stochastic Hormone Model

We have the same model

$$\frac{dR}{dt} = f(T) - b_1 R$$
$$\frac{dL}{dt} = g_1 R - b_2 L$$
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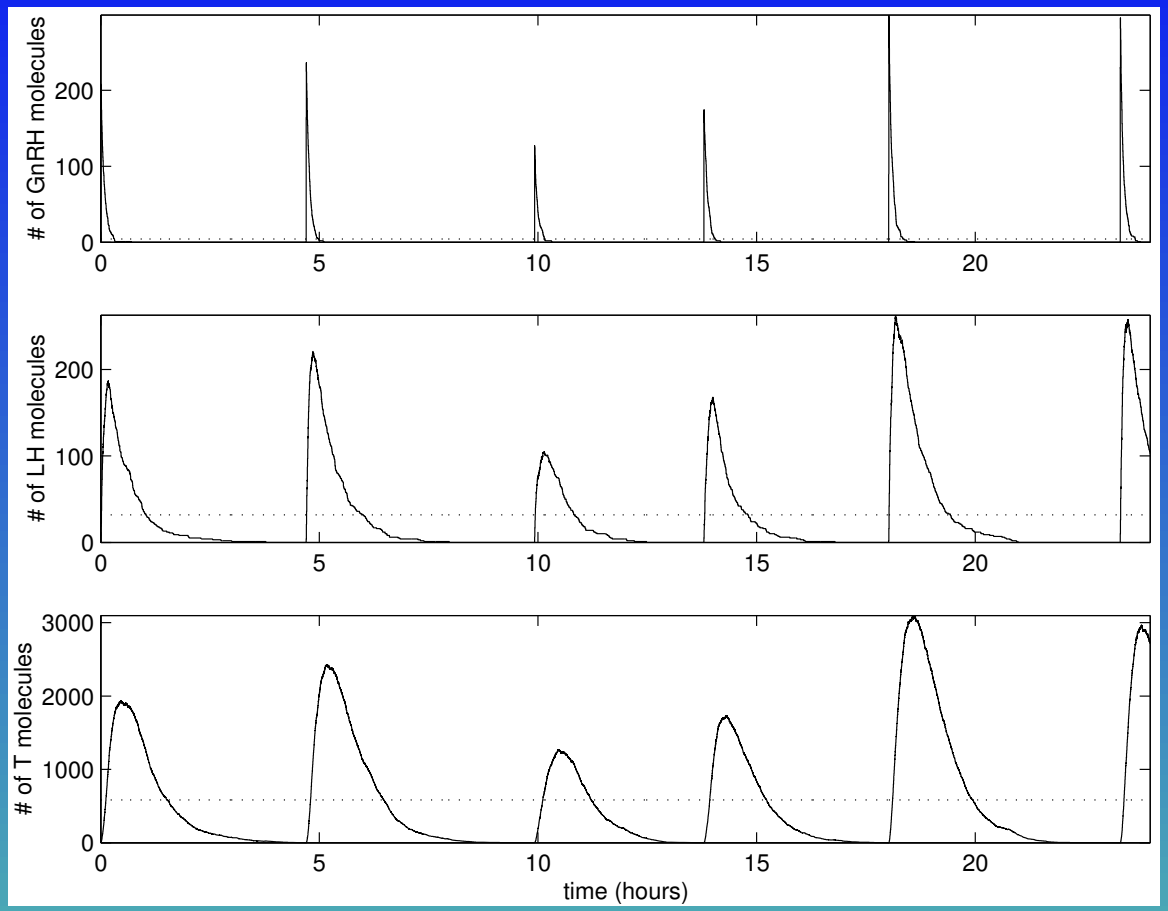
where

$$f(T) = \frac{A}{K+T},$$

but now

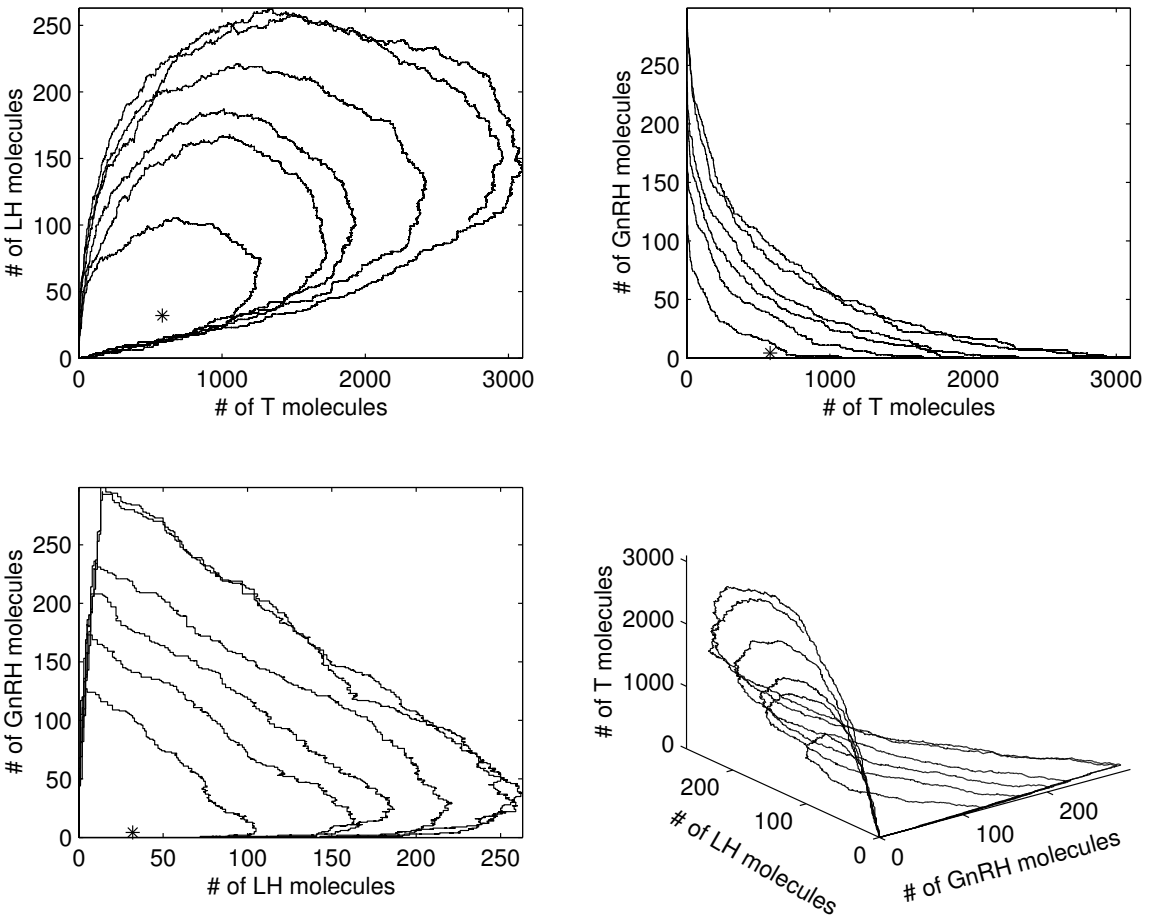
$$a_1 = \frac{A}{K + T(t)} \qquad a_2 = b_1 R(t)$$
$$a_3 = g_1 R(t) \qquad a_4 = b_2 L(t)$$
$$a_5 = g_2 L(t) \qquad a_6 = b_3 T(t).$$

A Stochastic Simulation



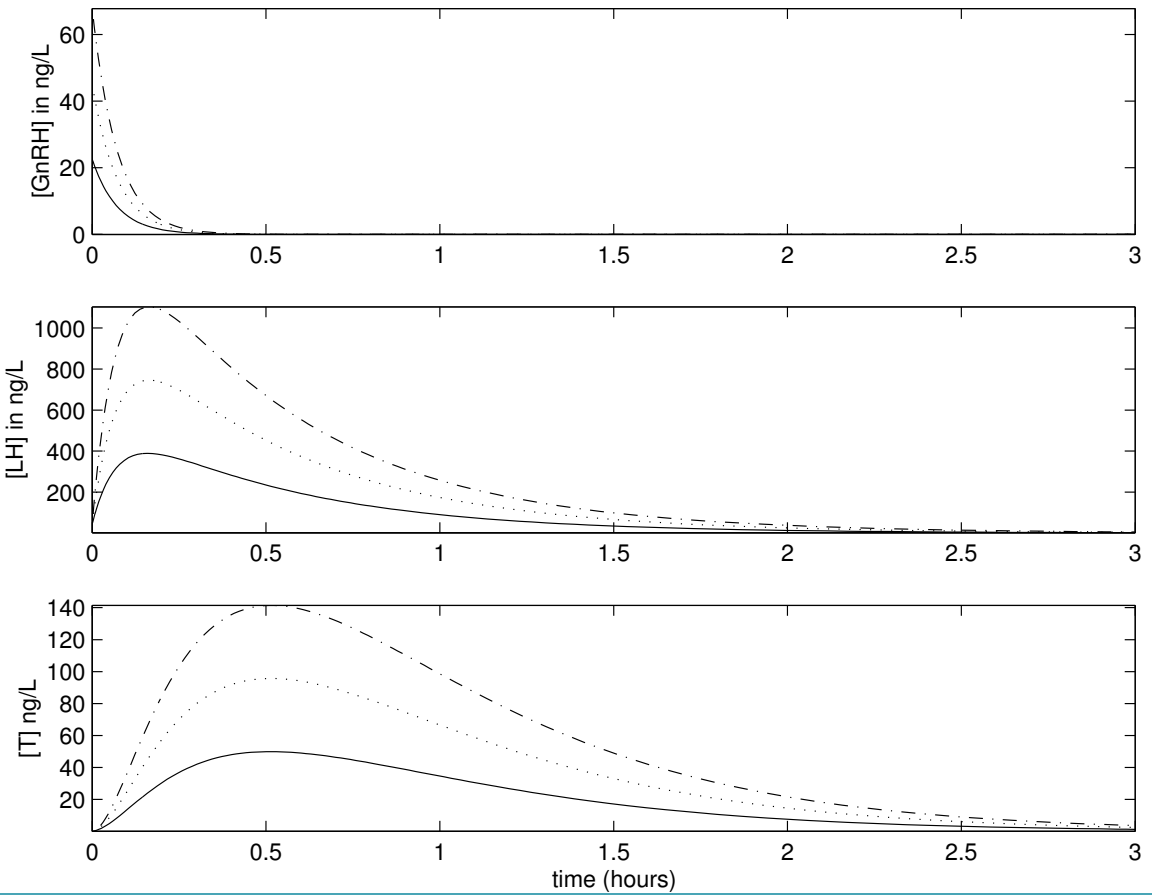
Simulation of hormone secretion for physical parameter values, $A = 10^{-4}$, $K = 10^{-7}$, $b_1 = 0.23$, $b_2 = 0.032$, $b_3 = 0.046$, $g_1 = 0.2618$, and $g_2 = 0.9015$. Average number of molecules are represented by dashed lines; average R is 4.20, average L is 31.90, and average T is 583.44. Volume is 10pL.

A Stochastic Simulation



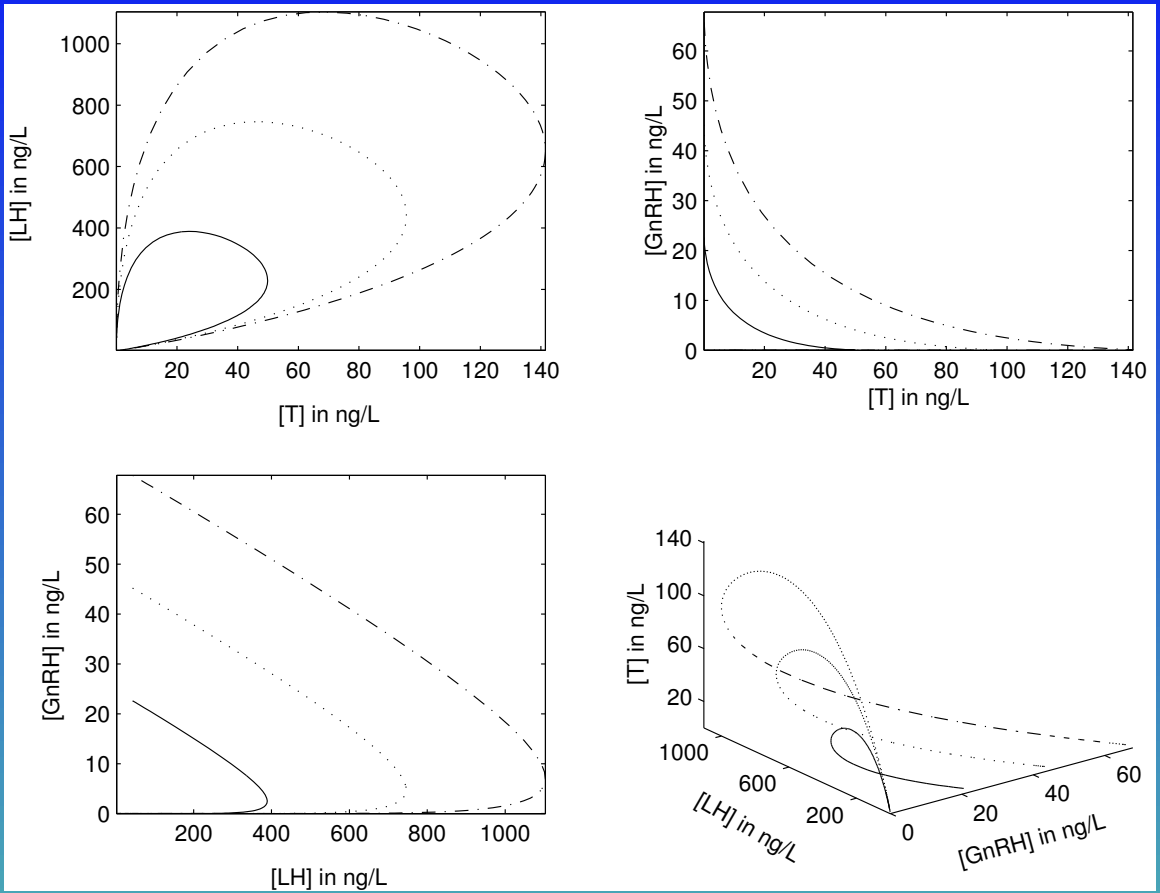
Two dimensional projections and three dimensional plot of simulation trajectory for the physical parameter values, $A = 10^{-4}$, $K = 10^{-7}$, $b_1 = 0.23$, $b_2 = 0.032$, $b_3 = 0.046$, $g_1 = 0.2618$, and $g_2 = 0.9015$. Average number of molecules are represented by asterisks; average R is 4.20, average L is 31.90, and average T is 583.44. Volume is 10pL.

Deterministic Model Trajectories



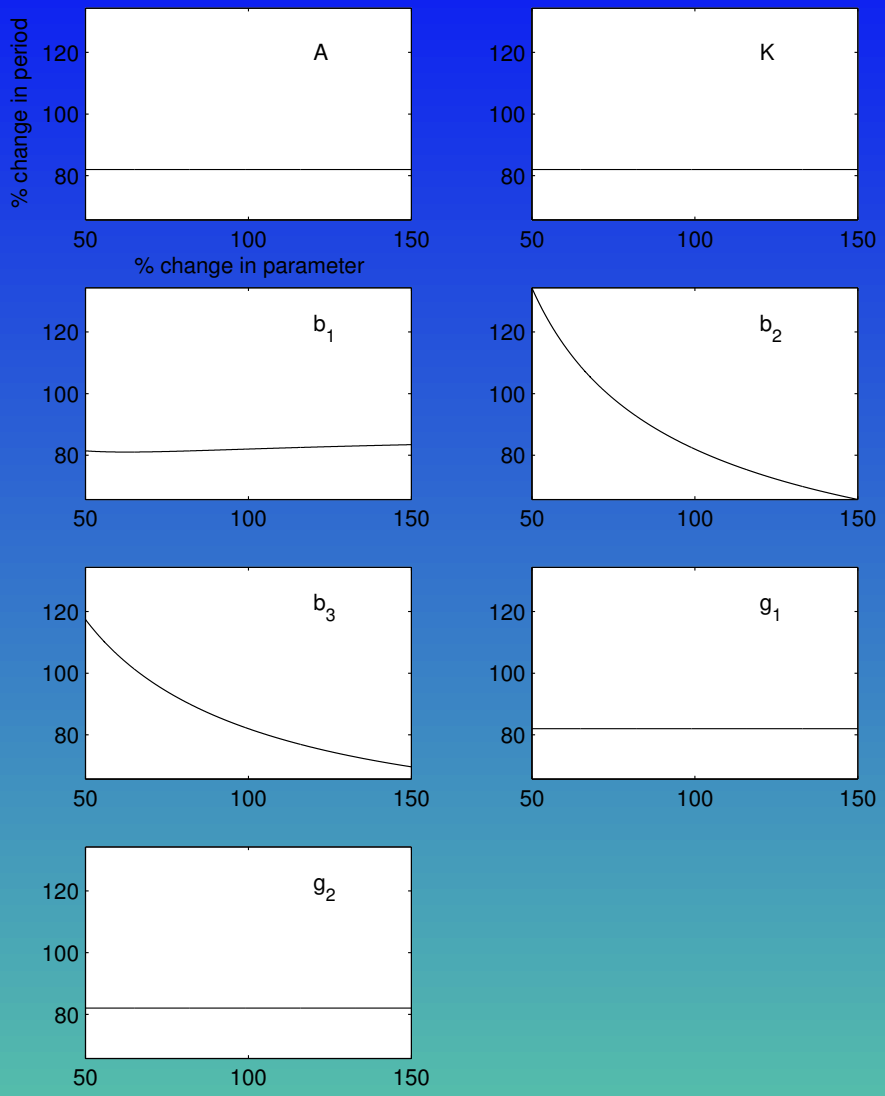
Solution trajectories for the deterministic hormone secretion model using parameter values equivalent to those used in the stochastic simulation. Initial conditions used for $GnRH$ are 22.61 ng/L (100 molecules / 10 pL) for solid curve, 45.21 ng/L (200 molecules / 10 pL) the dotted curve, and 67.82 ng/L (300 molecules / 10 pL) the dashed-dotted curve. Those for LH and T are 43.17 ng/L (10 molecules / 10 pL) and 0.05 ng/L (1 molecule / 10 pL), respectively.

Deterministic Model Trajectories



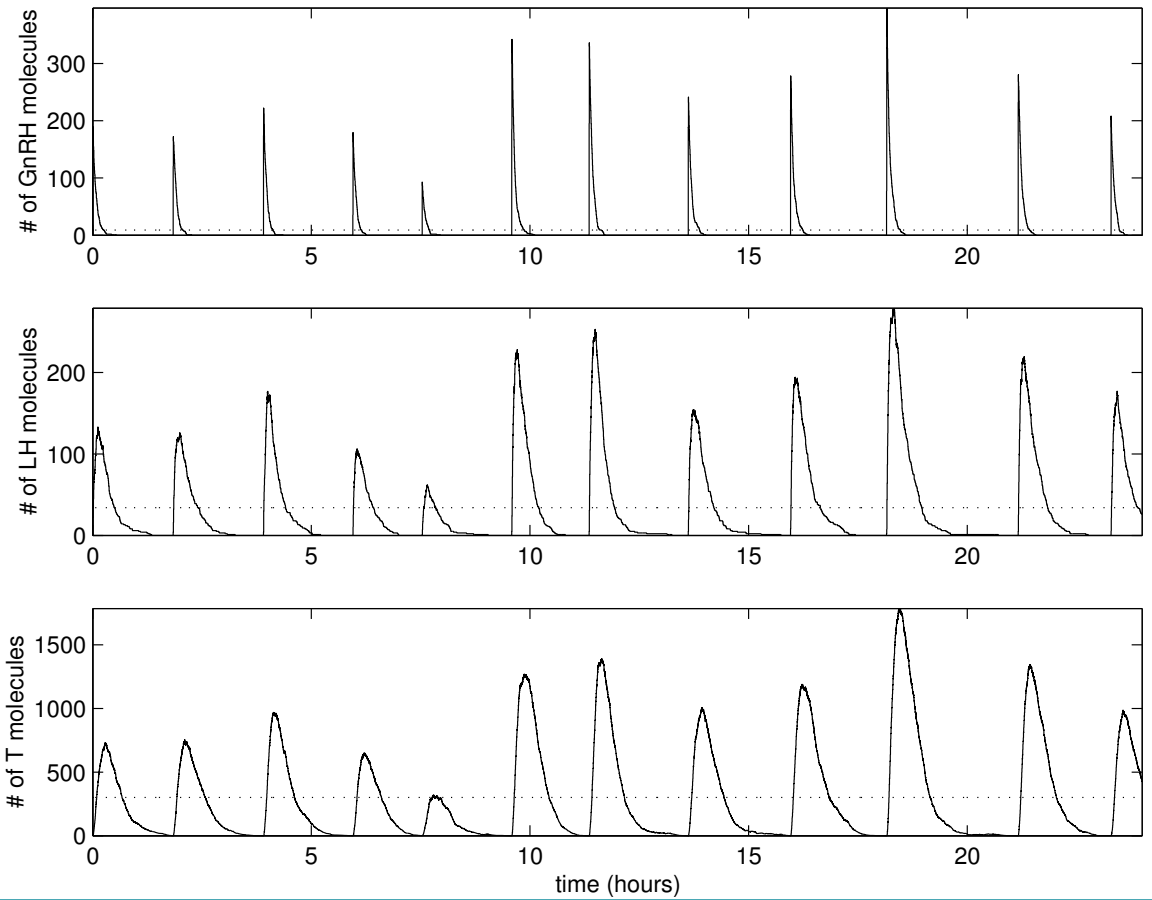
Two dimensional projections and three dimensional plot of the deterministic hormone secretion model trajectories using converted parameter values equivalent to those used in the stochastic simulation.

Period Sensitivity Analysis



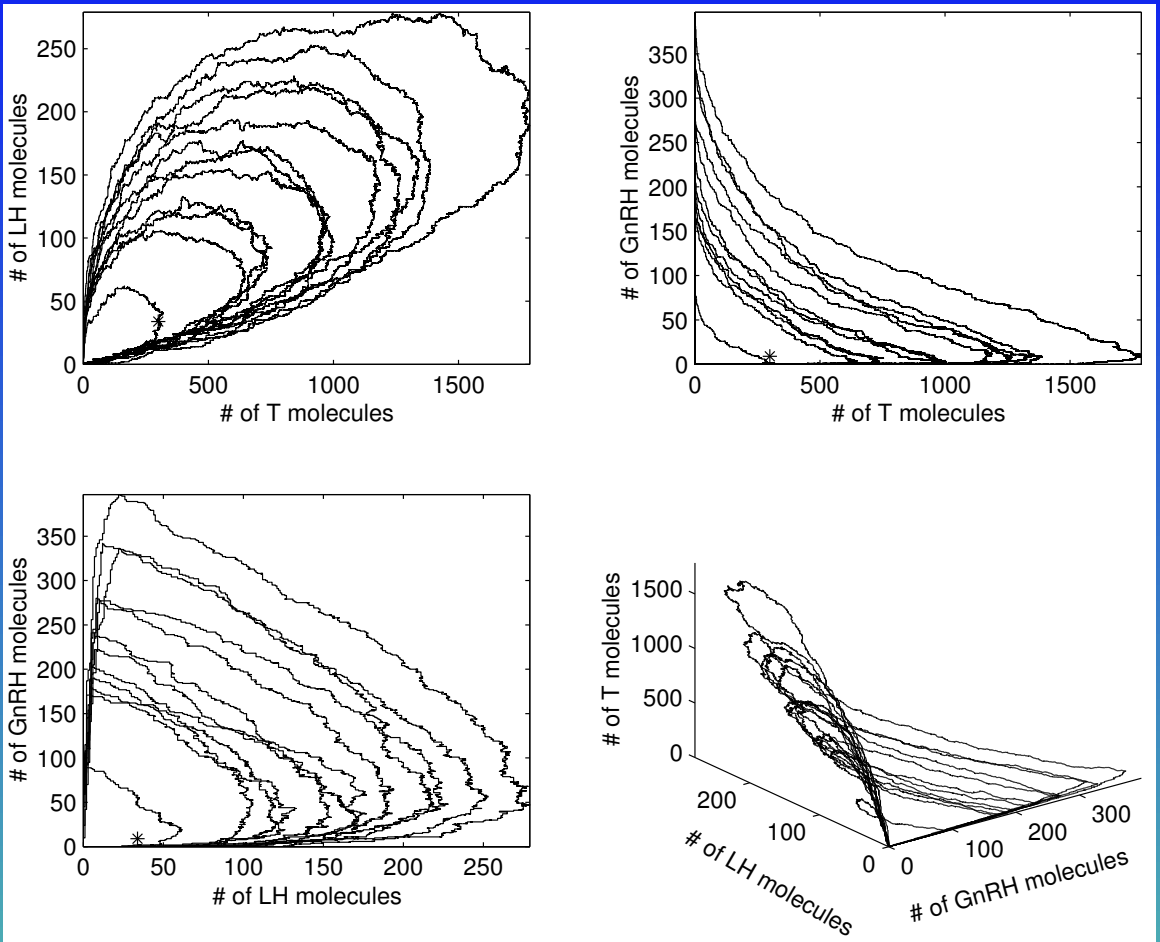
Initial eigenvalues were $\lambda_1 = -0.2385$ and $\lambda_{2,3} = -0.0347 \pm 0.0404i$.

A Stochastic Simulation



Simulation of hormone secretion for parameter values, $A = 10^{-4}$, $K = 10^{-7}$, $b_1 = 0.23$, $b_2 = 0.07$, $b_3 = 0.1$, $g_1 = 0.2618$, and $g_2 = 0.9015$. Average number of molecules are represented by dashed lines; average R is 9.09, average L is 33.92, and average T is 300.07. Volume is 10pL.

A Stochastic Simulation



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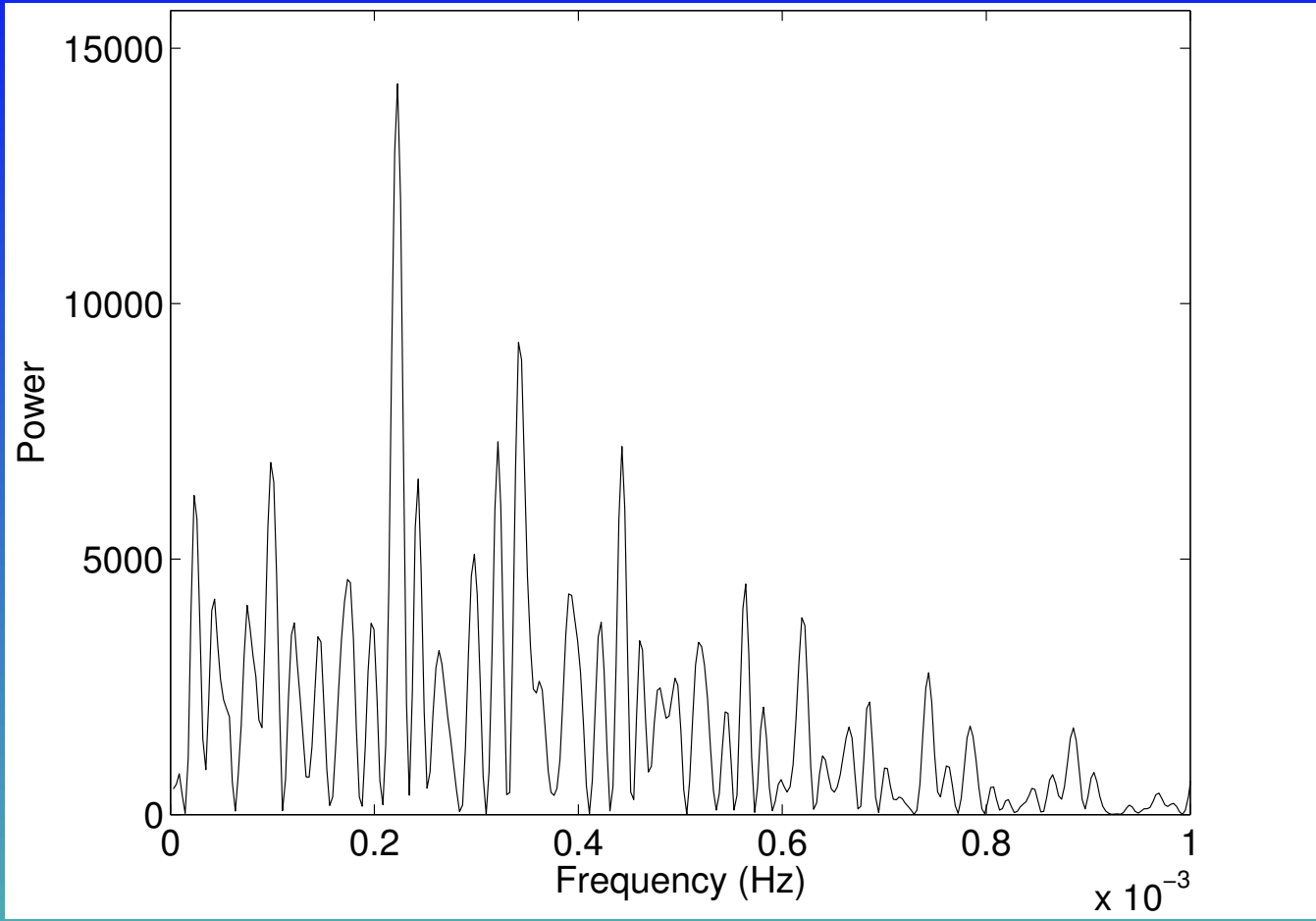
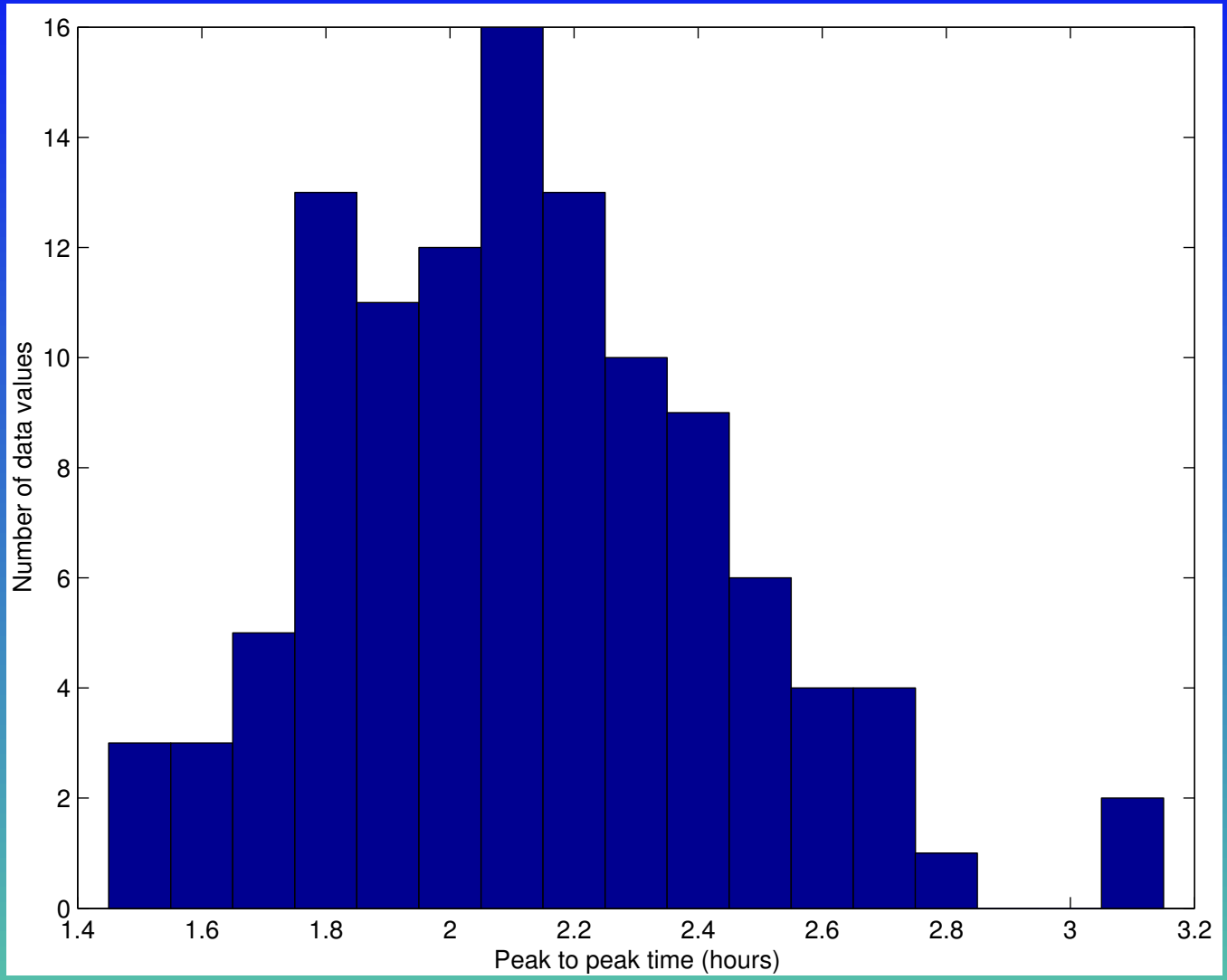
Lomb Spectral Analysis



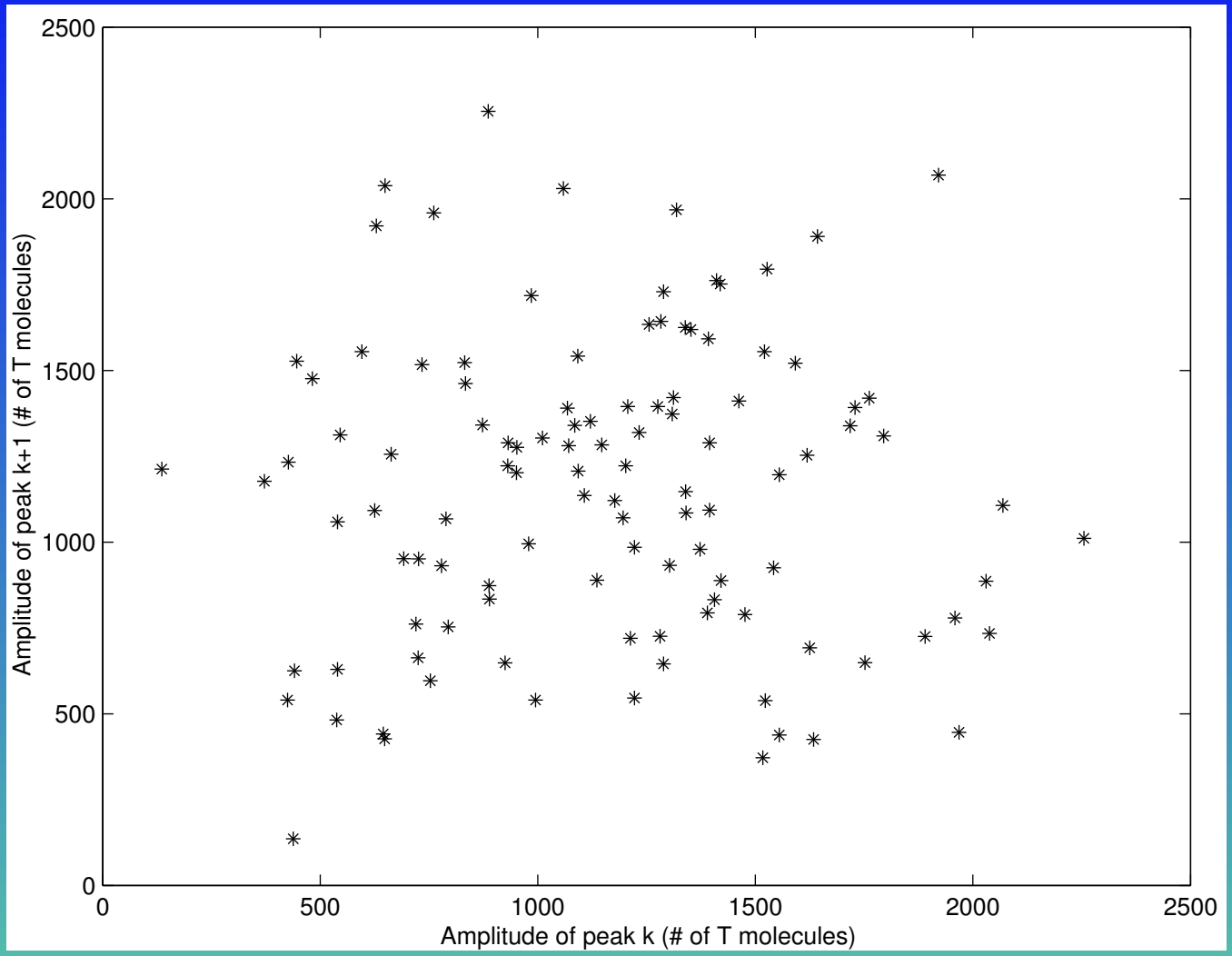
The largest peak corresponds to a frequency of 2.3429×10^{-4} Hz, which corresponds to a period of 1.2 hours.

Peak-to-Peak Time Histogram



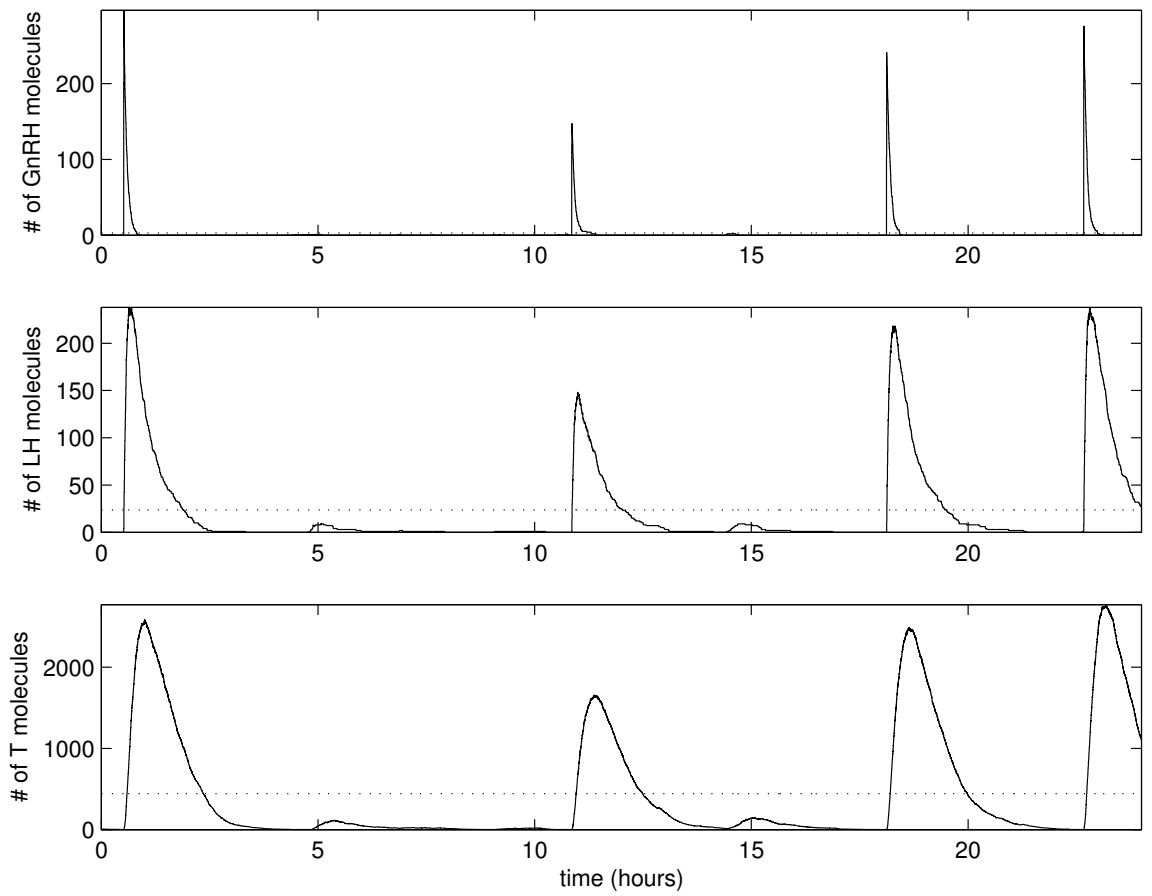
Histogram of peak-to-peak time intervals for the hormone secretion pathway over a 10 day time period.

Peak-to-Peak Amplitude



Peak-to-peak return map illustrating the amplitude of one pulse relative to that of the preceding pulse.

The Switching Behavior



Simulation to illustrate the the switching behavior. Parameter values are $A = 10^{-1}$, $K = 10^{-4}$, $b_1 = 0.23$, $b_2 = 0.032$, $b_3 = 0.046$, $g_1 = 0.2618$, and $g_2 = 0.9015$.

Approximation Methods

- Gibson and Bruck (2000) proposed an approximation for systems in which some reactions occur much more often than others by reducing the number of random variables simulated.
- Gillespie (2001) introduced the τ-leap methods that make larger time steps and allow more events to occur within those steps as long as changes in the event probabilities stay within some tolerance.
- Burrage and Tian (2003) attempted to simulate continuous-time, continuous-state, stochastic-approximation, models driven by Wiener noise by introducing the framework of Poisson–Runge–Kutta methods.
- Turner, Schnell, and Burrage (2004) included fluctuations caused by the structural organisation of the cytoplasm and the limited diffusion of molecules due to macromolecular crowding.
- Burrage, Tian, and Burrage (2004) used multi-scale methods to incorporate the quasi-steady-state assumption with slow, intermediate, and fast reactions.

Conclusions

- By approaching the hormone model from a different physical basis we saw how intrinsic fluctuations can incite oscillations for low numbers of molecules by way of a switching behavior.
- Even though the deterministic model has a globally stable fixed point, the stochastic model was able to capture the pulsatile behavior of the blood hormone levels.
- When we are interested in the effects of intrinsic fluctuations and are not able to obtain analytic results, we can rely on simulation methods such as the Gillespie algorithm.
- These algorithms provide a more realistic representation of a system than the deterministic, mass-action equations.
- The Gillespie algorithm is easy to implement, but can be computationally demanding. However, approximation methods can be used to speed up the computations.

Future Work

- Do a Poincaré-map-like analysis of the oscillations we see with the stochastic model and study the resulting distribution.
- We are interested in finding out more about the relationship between the deterministic and stochastic frequencies.
- Further details can be incorporated into the model, such as basal hormone secretion, temporal shifts, and additional negative feedback relationships in the signaling pathway.

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